A molecular signature for purified definitive endoderm guides differentiation and isolation of endoderm from mouse and human embryonic stem cells.

Journal: Stem Cells Dev

Publication Year: 2012

Authors: P Wang, K D McKnight, D J Wong, R T Rodriguez, T Sugiyama, X Gu, A Ghodasara, K Qu, H Y Chang, S Kim

PubMed link: 22236333

Funding Grants: Noncoding RNAs in Cell Fate Determination

Public Summary:
Embryonic definitive endoderm generates the epithelial compartment of vital organs like liver, pancreas and intestine. However, purification of definitive endoderm in mammals has not been achieved, limiting the molecular 'definition' of endoderm, and hindering our understanding of definitive endoderm development and attempts to produce endoderm from sources like ES cells. Here, we use mouse and human embryonic stem cells to improve definitive endoderm development in vitro, which guides further use of ES cells for tissue replacement.

Scientific Abstract:
Embryonic definitive endoderm generates the epithelial compartment of vital organs like liver, pancreas and intestine. However, purification of definitive endoderm in mammals has not been achieved, limiting the molecular 'definition' of endoderm, and hindering our understanding of definitive endoderm development and attempts to produce endoderm from sources like ES cells. Here, we describe purification of mouse definitive endoderm using fluorescence-activated cell sorting (FACS) and mice harboring a transgene encoding enhanced green fluorescent protein (eGFP) inserted into the Sox17 locus, which is expressed in embryonic endoderm. Comparison of patterns of signaling pathway activation in native mouse definitive endoderm and endoderm-like cells generated from ES cells produced novel culture modifications that generated Sox17-eGFP+ progeny whose gene expression resembled definitive endoderm more closely than achieved with standard methods. These studies also produced new FACS methods for purifying definitive endoderm from non-transgenic mice and mouse ES cell cultures. Parallel studies of a new human SOX17-eGFP ES cell line allowed analysis of endoderm differentiation in vitro, leading to culture modifications that enhanced expression of an endoderm-like signature. This work should accelerate our understanding of mechanisms regulating definitive endoderm development in mice and humans, and guide further use of ES cells for tissue replacement.